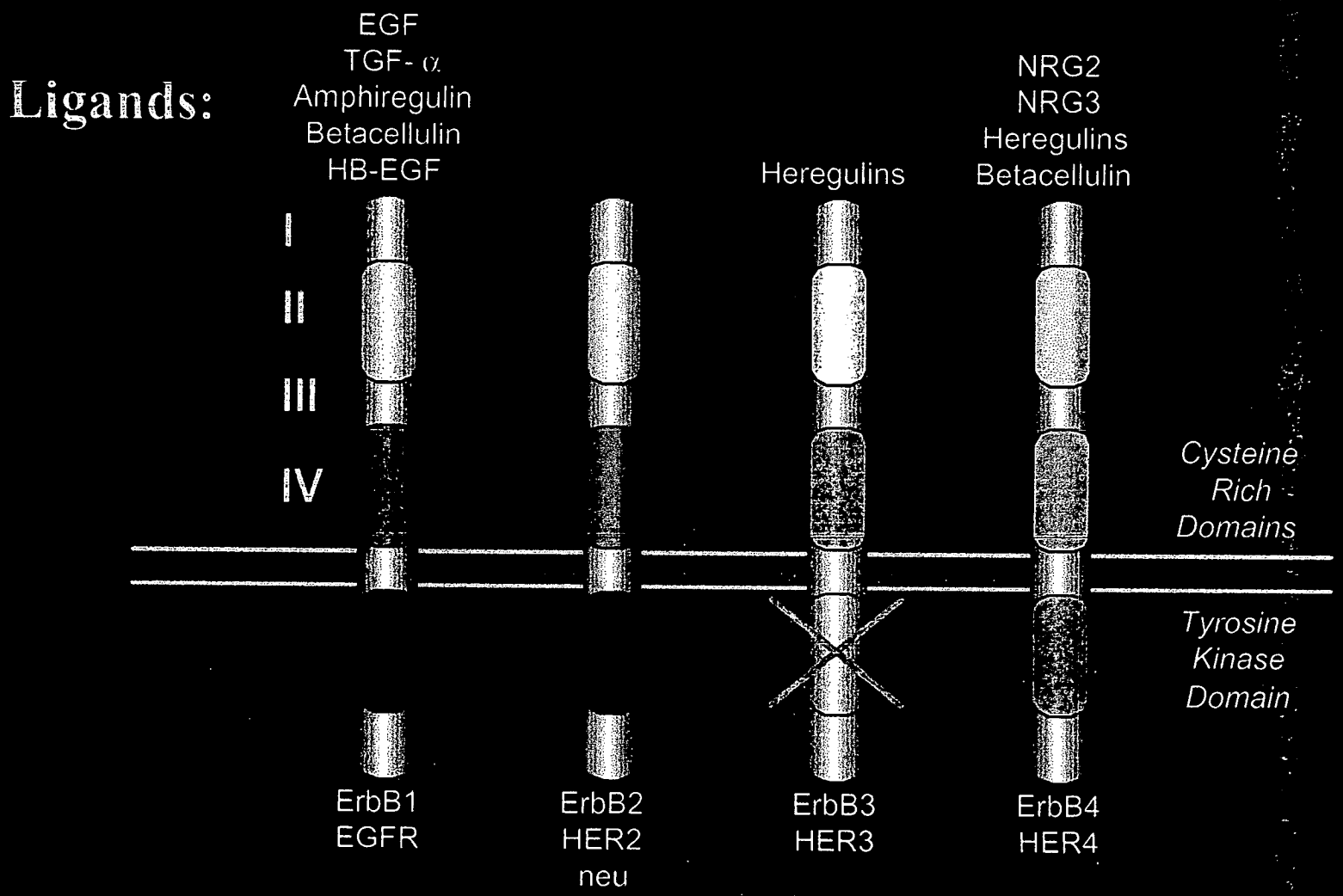
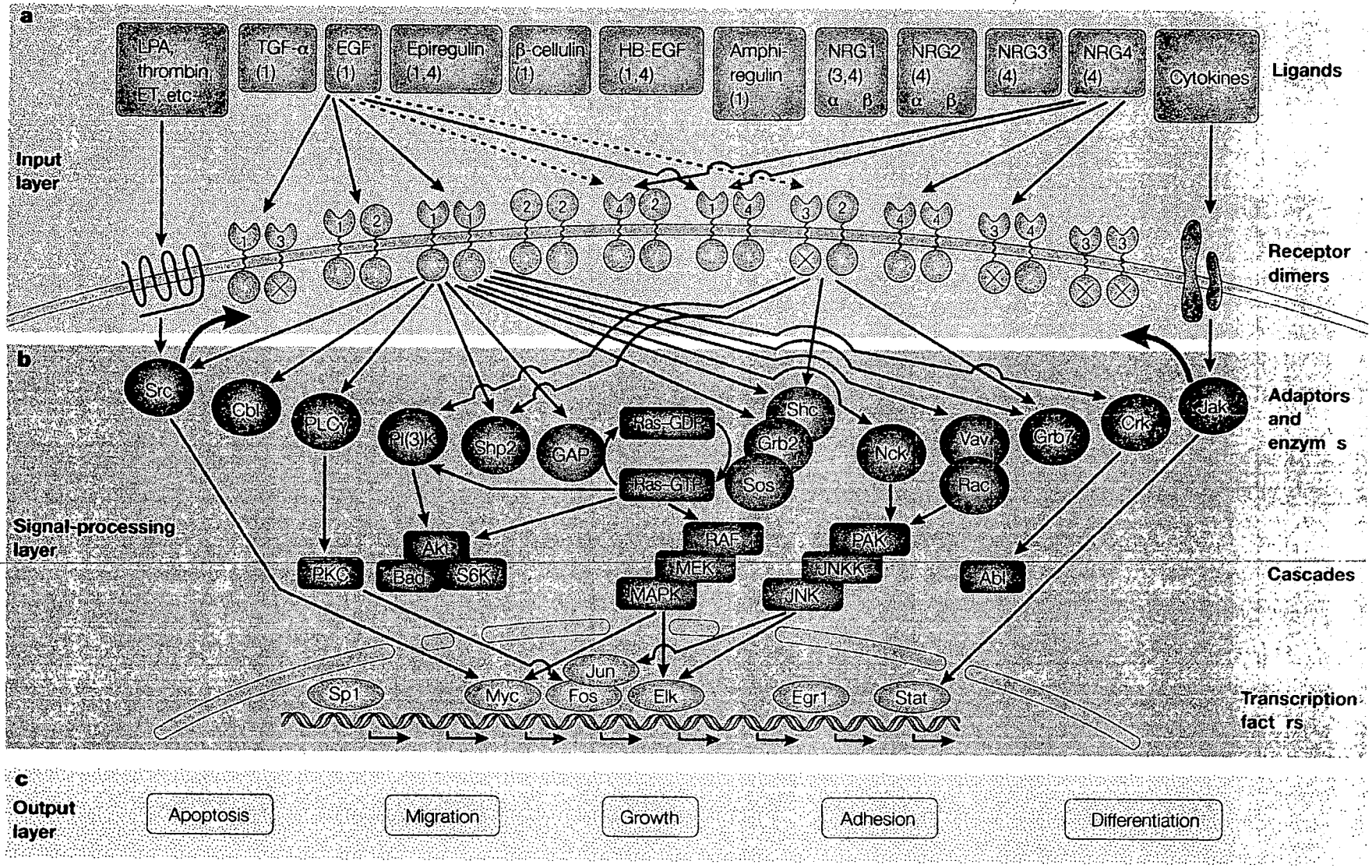
Evolution of the HER/ErbB Receptor System

Worms:	1 Ligand and 1 Receptor
Flies:	4 Ligands and 1 Receptor
Mammals:	12 Ligands and 4 Receptors

The HERs
A Dysfunctional Family of Receptors



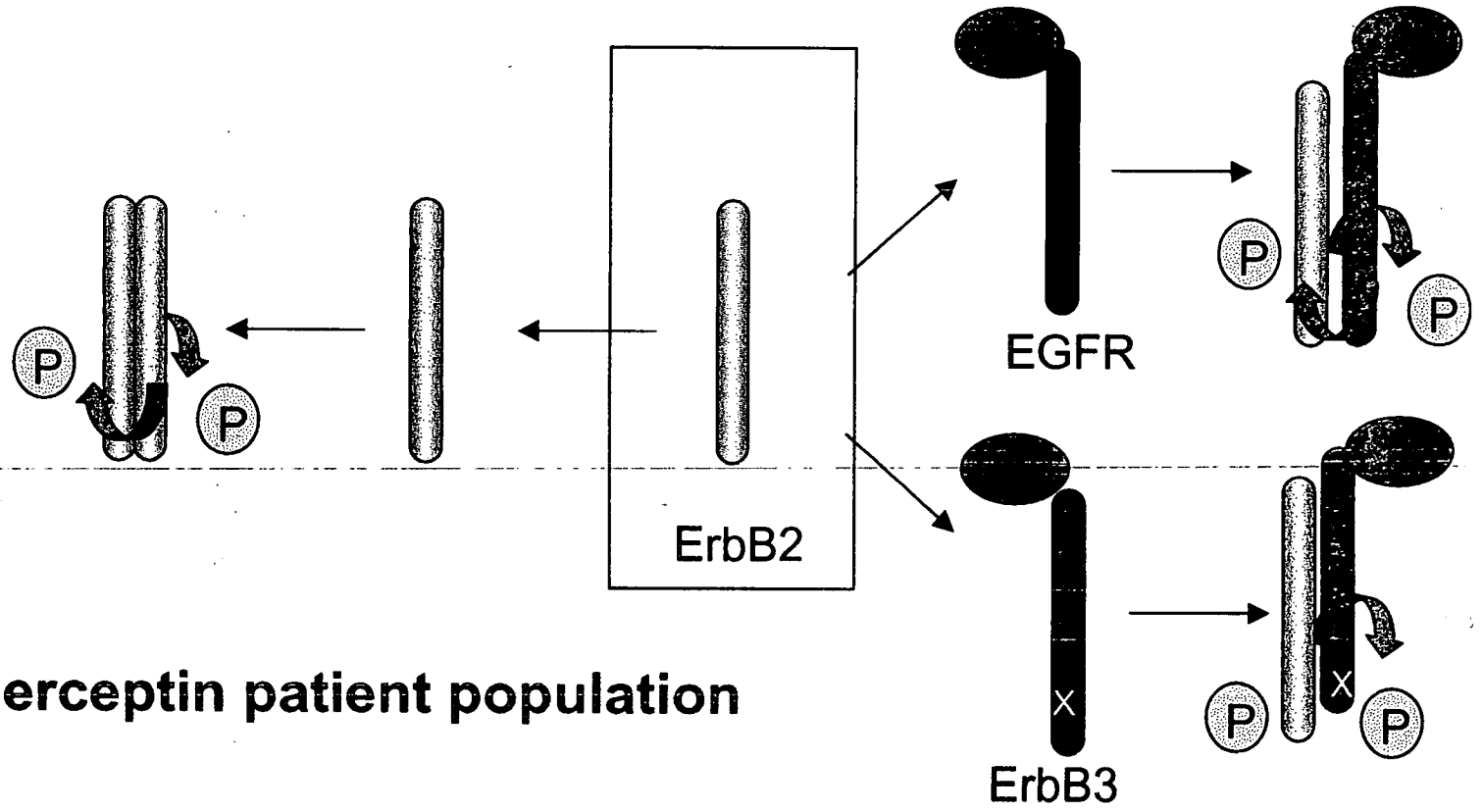


From Yarden & Sliwkowski. Nature Reviews MCB(2001)

HER2 Activation in Cancer

Ligand-independent (Amplified HER2 tumors)

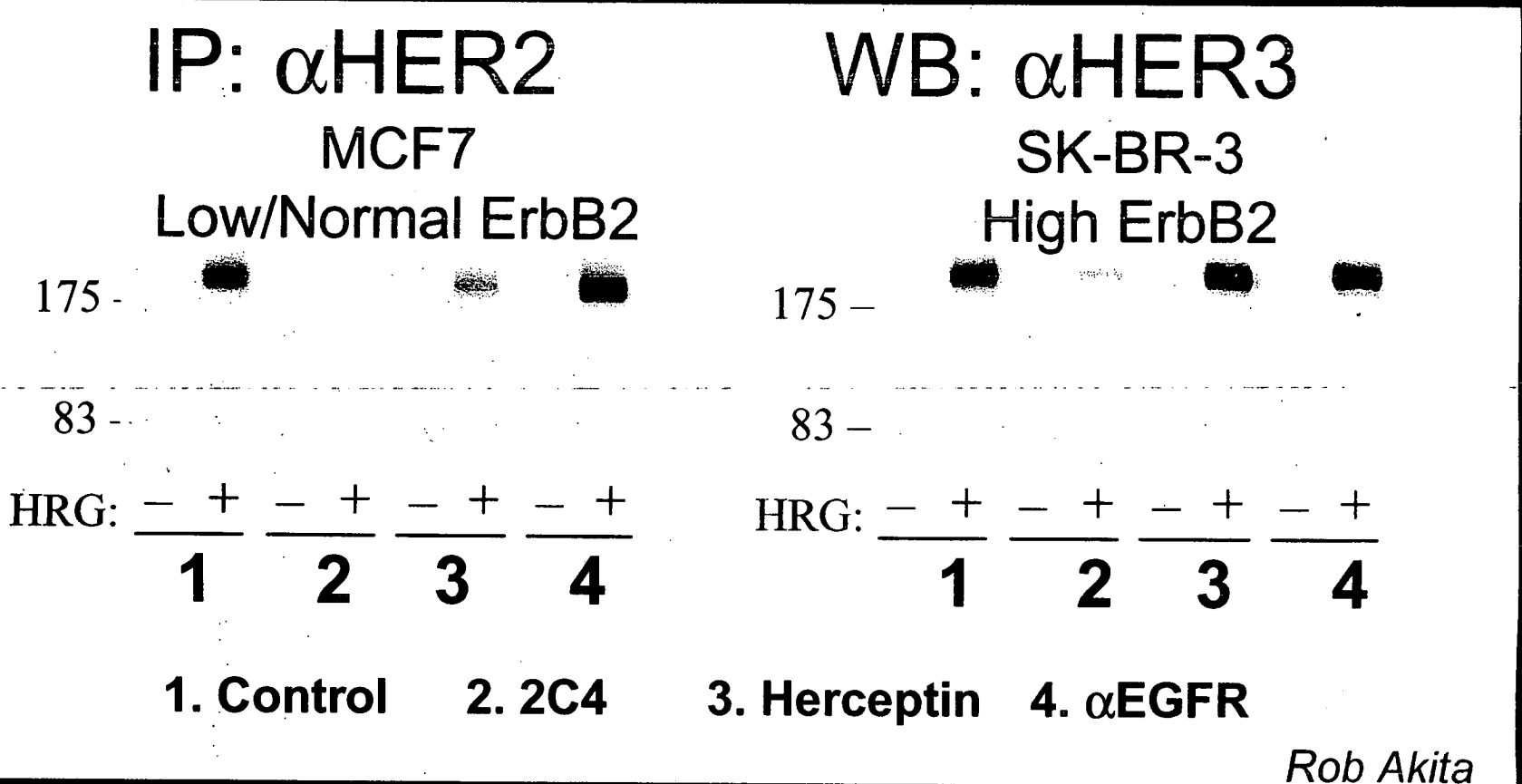
Ligand-dependent (Non-amplified HER2 tumors)



Herceptin patient population

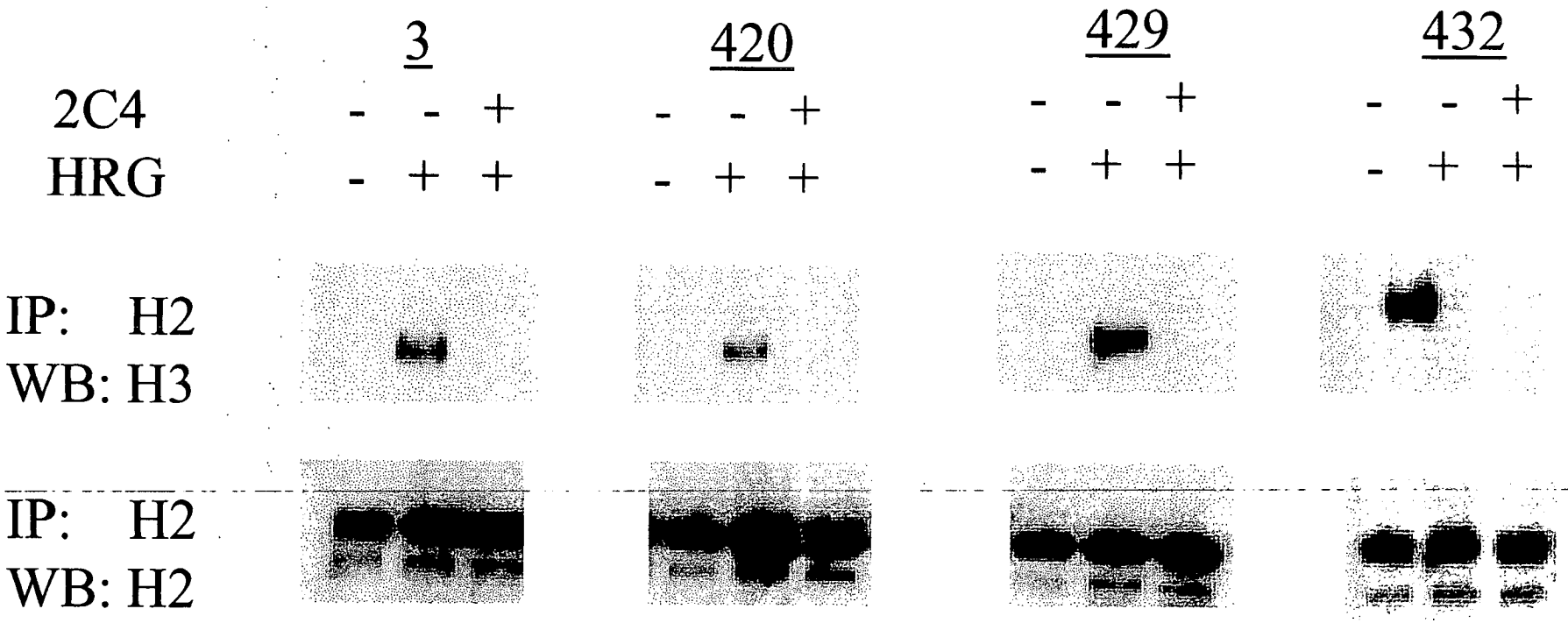
HER2 Associates with HER3 in a Heregulin-Dependent Manner

- 2C4 blocks ligand-dependent HER2-HER3 association, Herceptin does not.

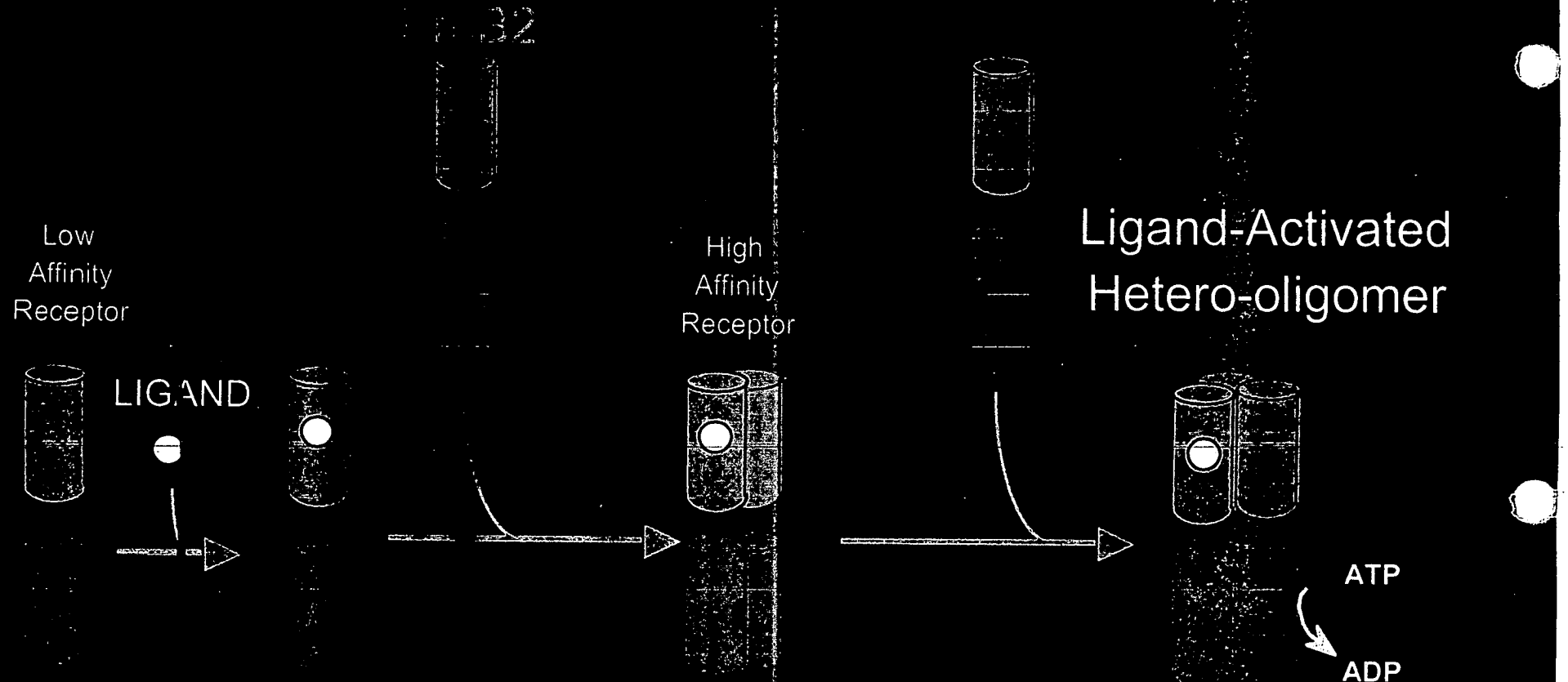


Rob Akita

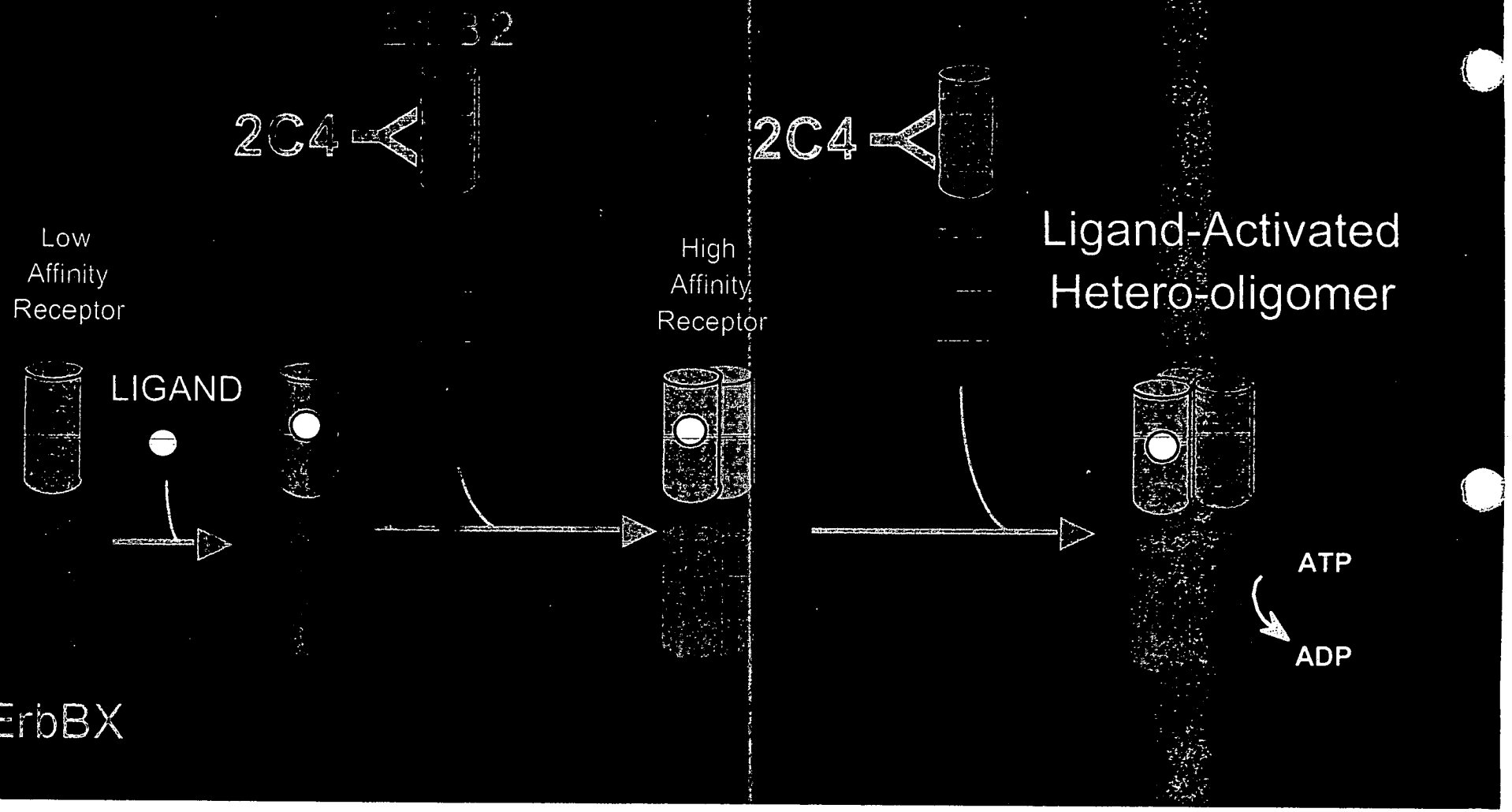
Ovarian Tumor Cell Lines



ErbB2 is recruited to ErbB3-HRG Complexes

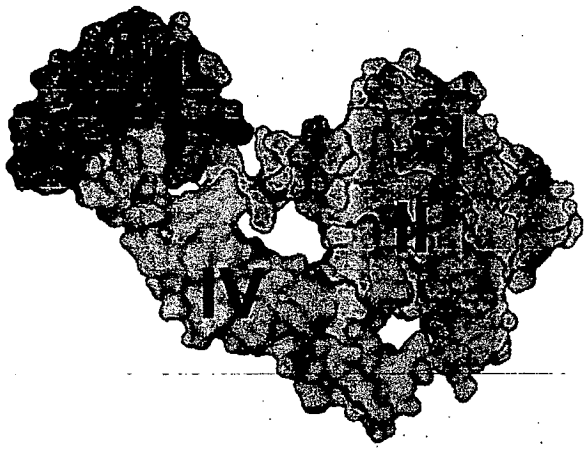


2C4 Disrupts Ligand-Dependent ErbB2 Signaling



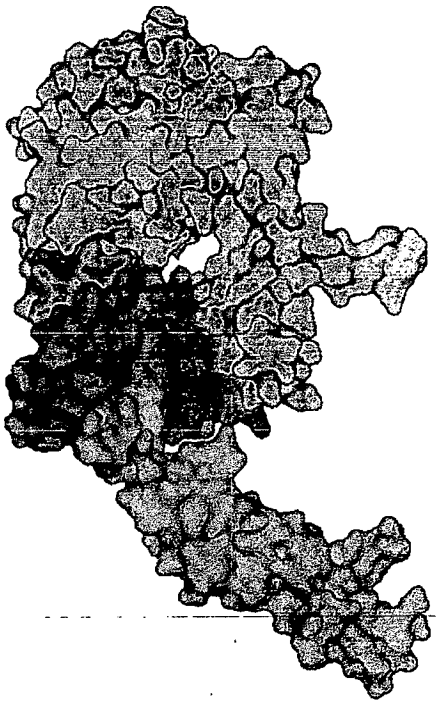
EGFR

Closed



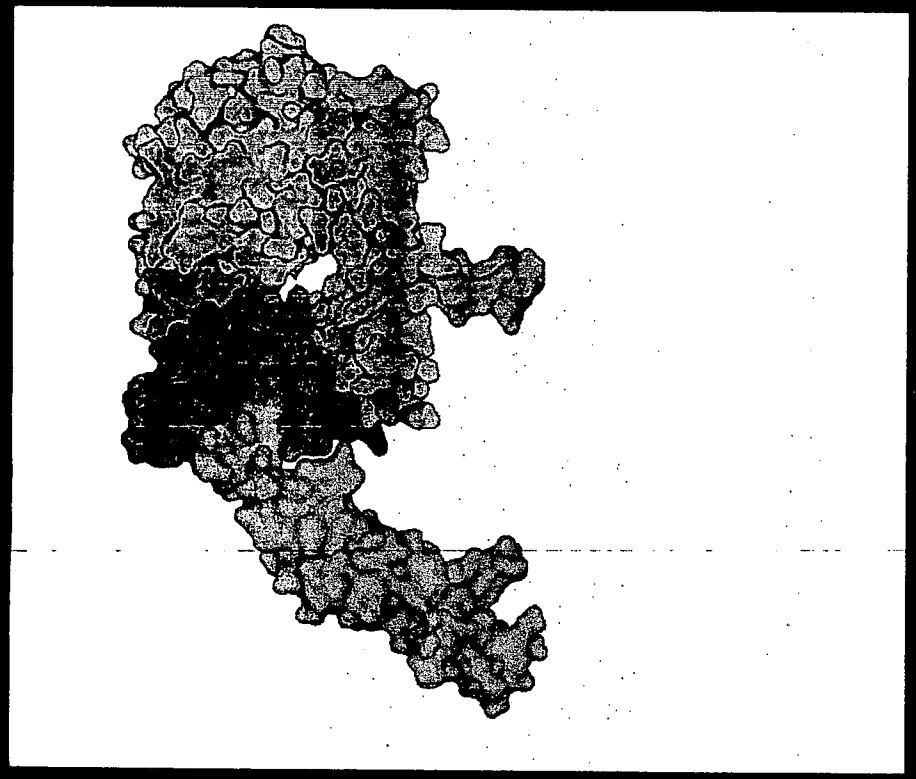
EGFR-EGF Complex

Open

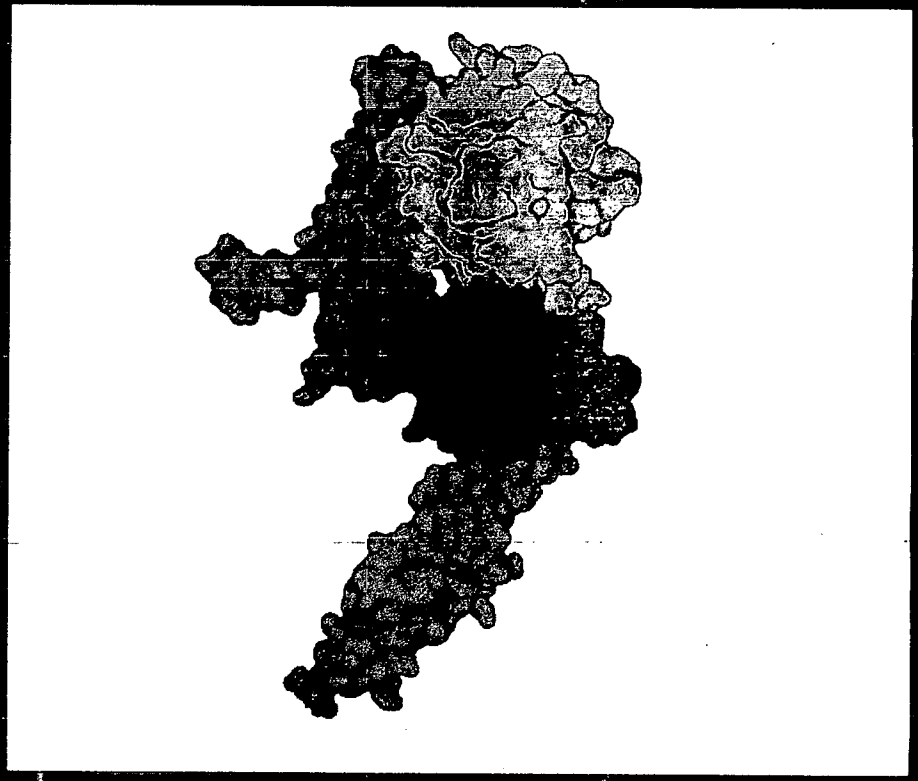


Ogiso et al. Cell (2002) 110: 775
Garret et al. Cell (2002) 110: 763
Ferguson et al. Mol Cell (2003) 11:507

EGFR-EGF Complex

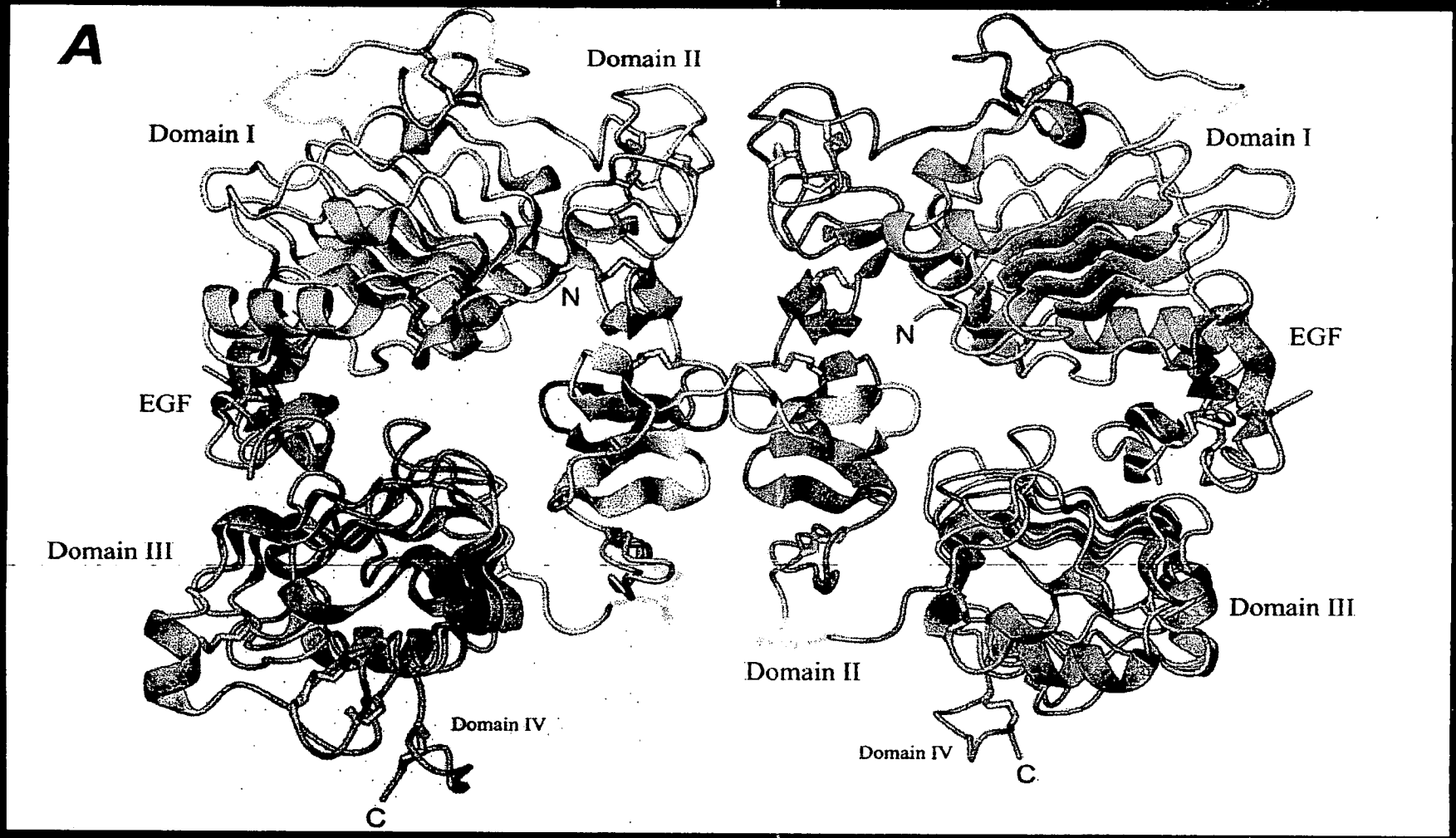


HER2



Cho et al. Nature 421:756.
Matt Franklin & Bart de Vos, Genentech

Receptor dimerization via a domain II handshake



Ogiso et al Cell (2002) 110: 775

Comparison of HER2-EGFR to EGFR-EGFR Complexes

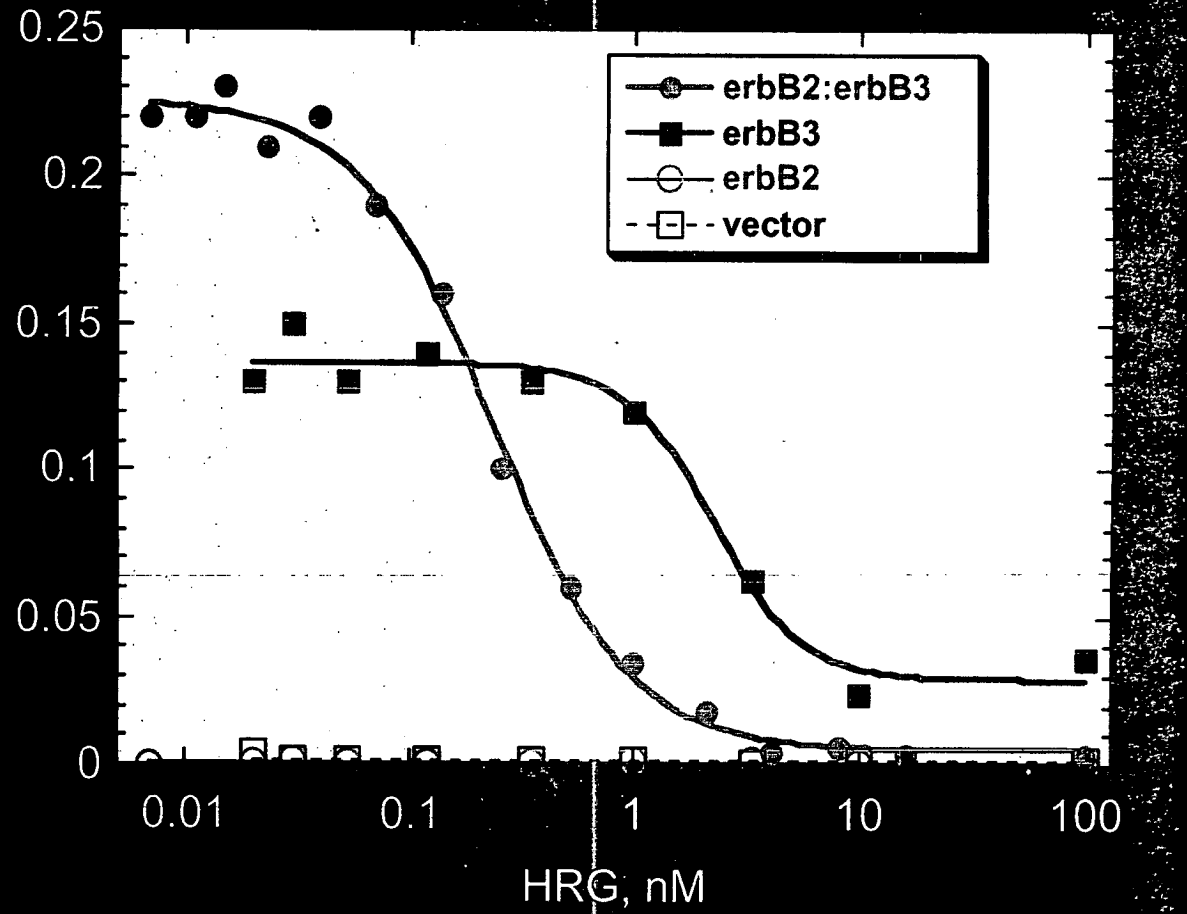
Characteristics of heterocomplexes :

- Higher affinity
- Decreased internalization rates
- Altered trafficking
- Diverse downstream signaling

HER2-HER3 Complex: A Paradigm for Efficient Molecular Collaboration

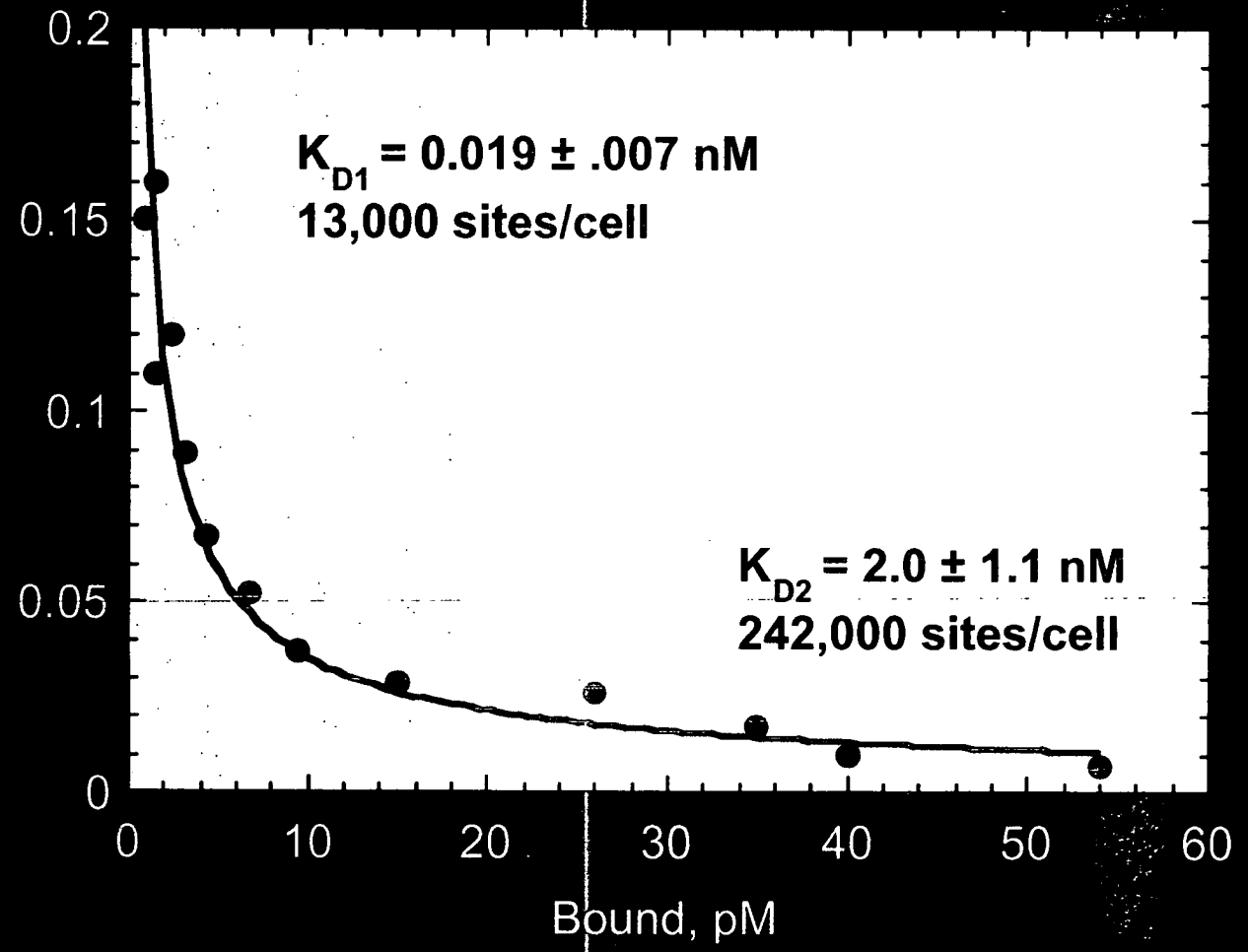
- Symbiotic relationship
  - Ligand-less HER2 and defective-kinase HER3.
- Most potent HER signaling complex.
- Efficiently activates both MAPK and PI3K signaling pathways.
  - HER2's active kinase
  - HER3 serves as a kinase substrate for HER2.
    - Multiple potential tyrosine phosphorylation sites.
    - Especially for PI3-kinase.
- Most active complex with regard to transformation potential.

Heregulin Binding to Cos Cell Transfectants



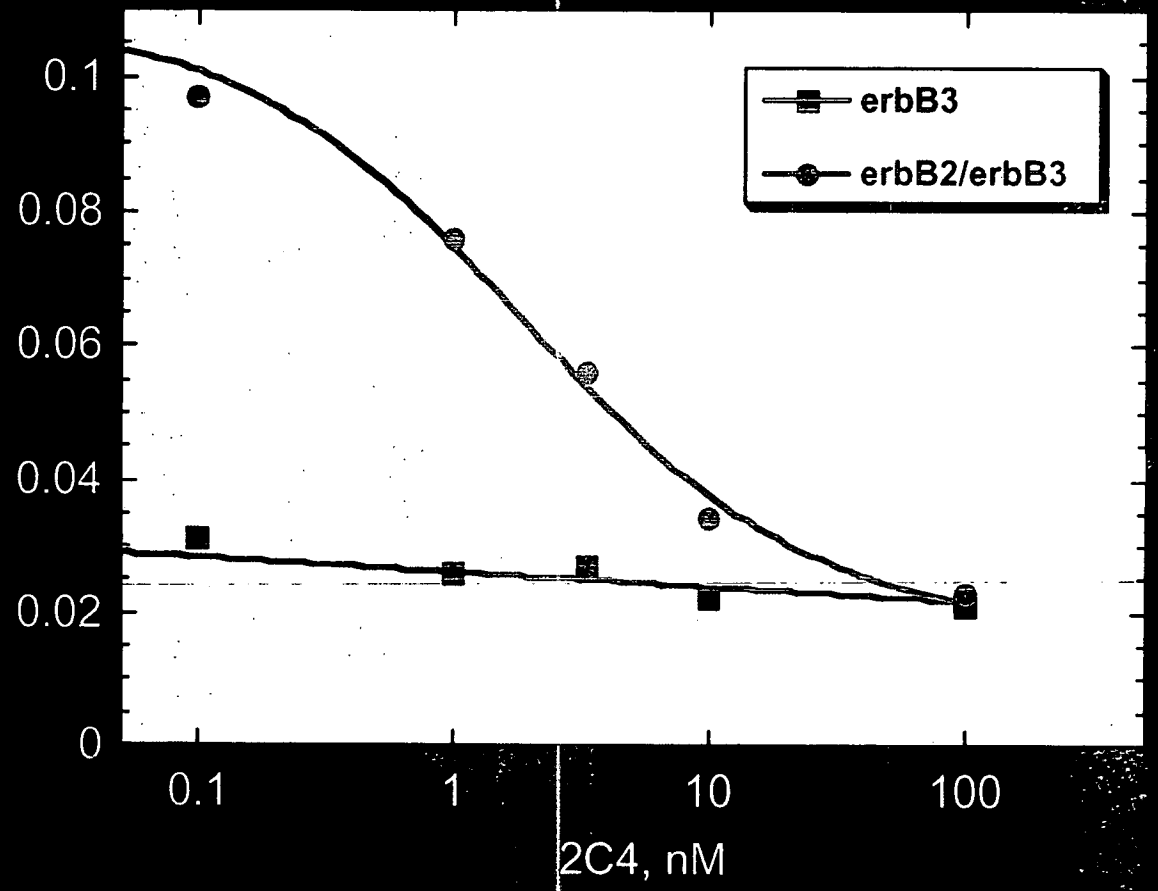
Gabriele Schaefer

Scatchard Analysis of Heregulin Binding to Cells Transfected with HER3 and HER2

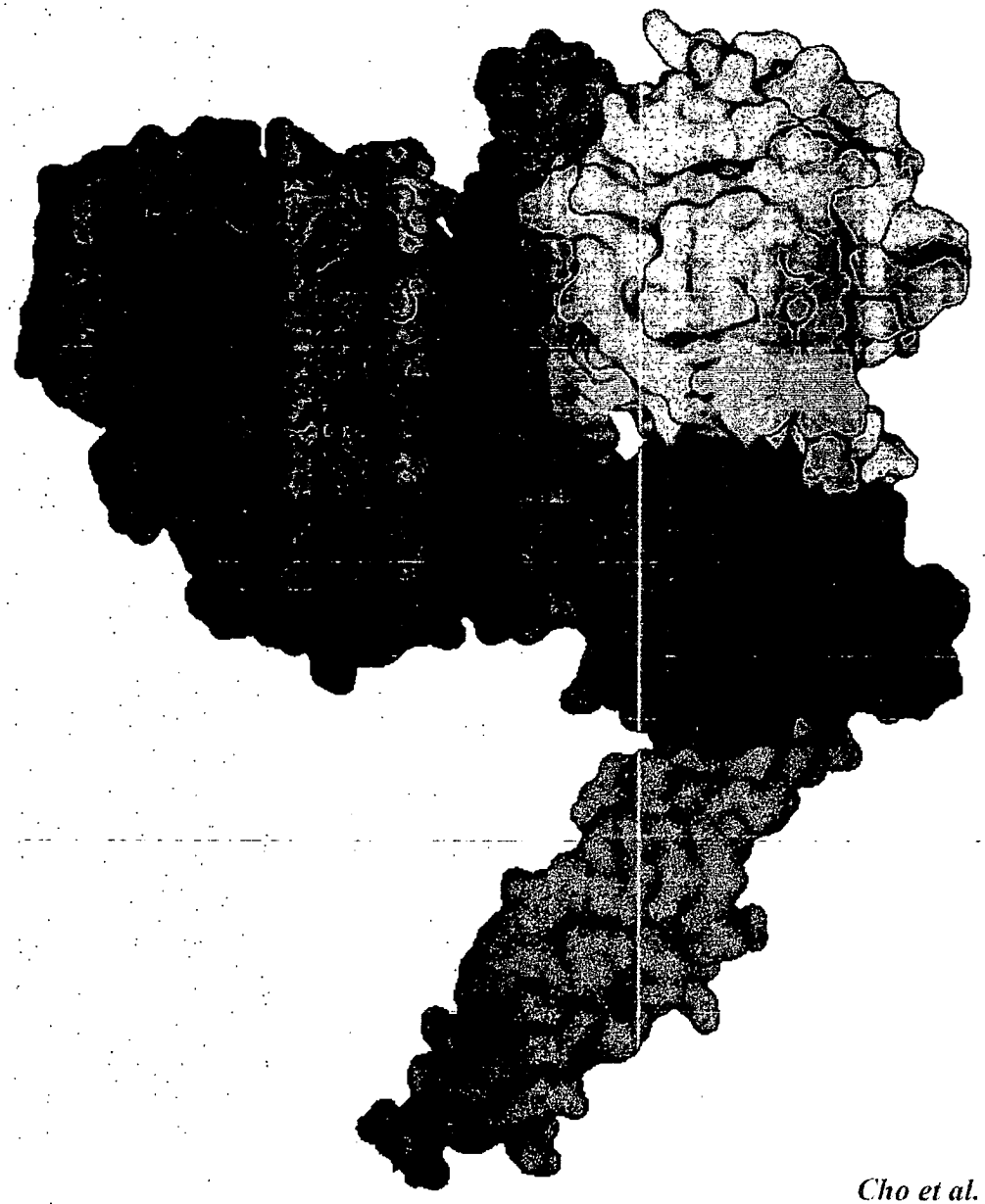


Gabriele Schaefer

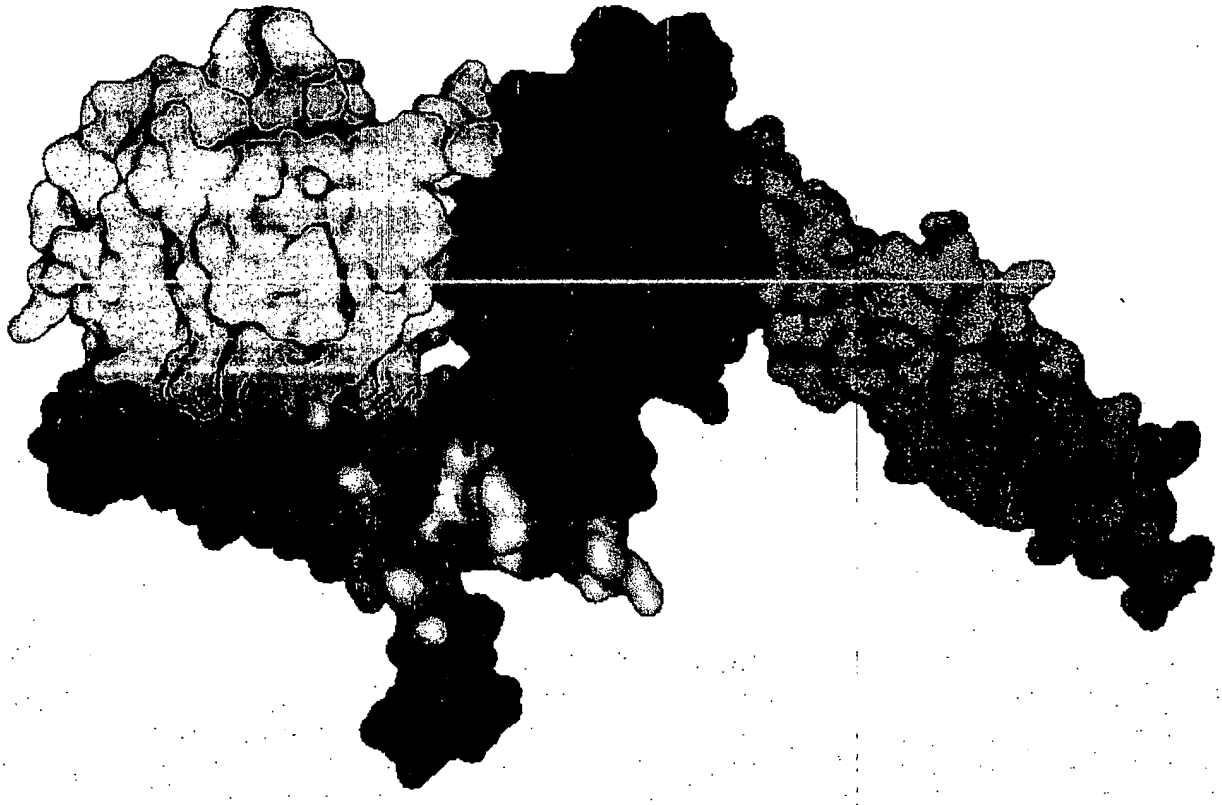
Inhibition of Heregulin Binding by 2C4 a Monoclonal Antibody to ErbB2



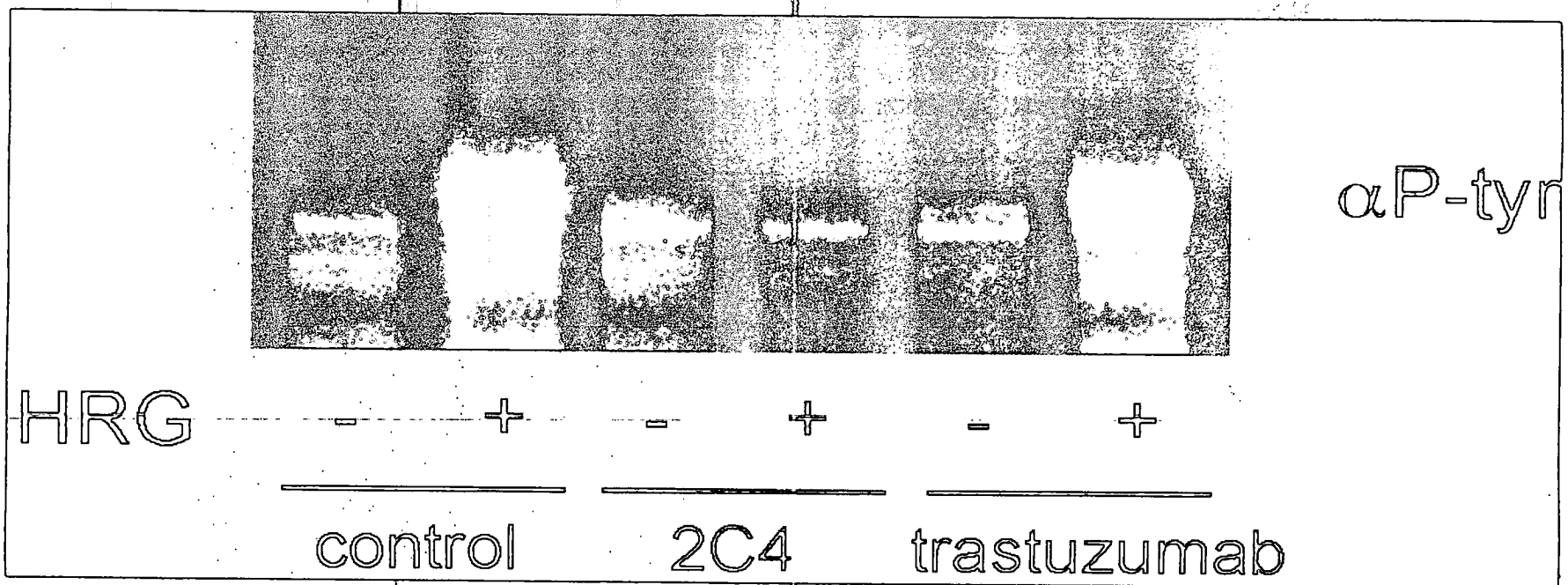
Gabriele Schaefer



Cho et al. Nature 421:756.
Matt Franklin & Bart de Vos, Genentech

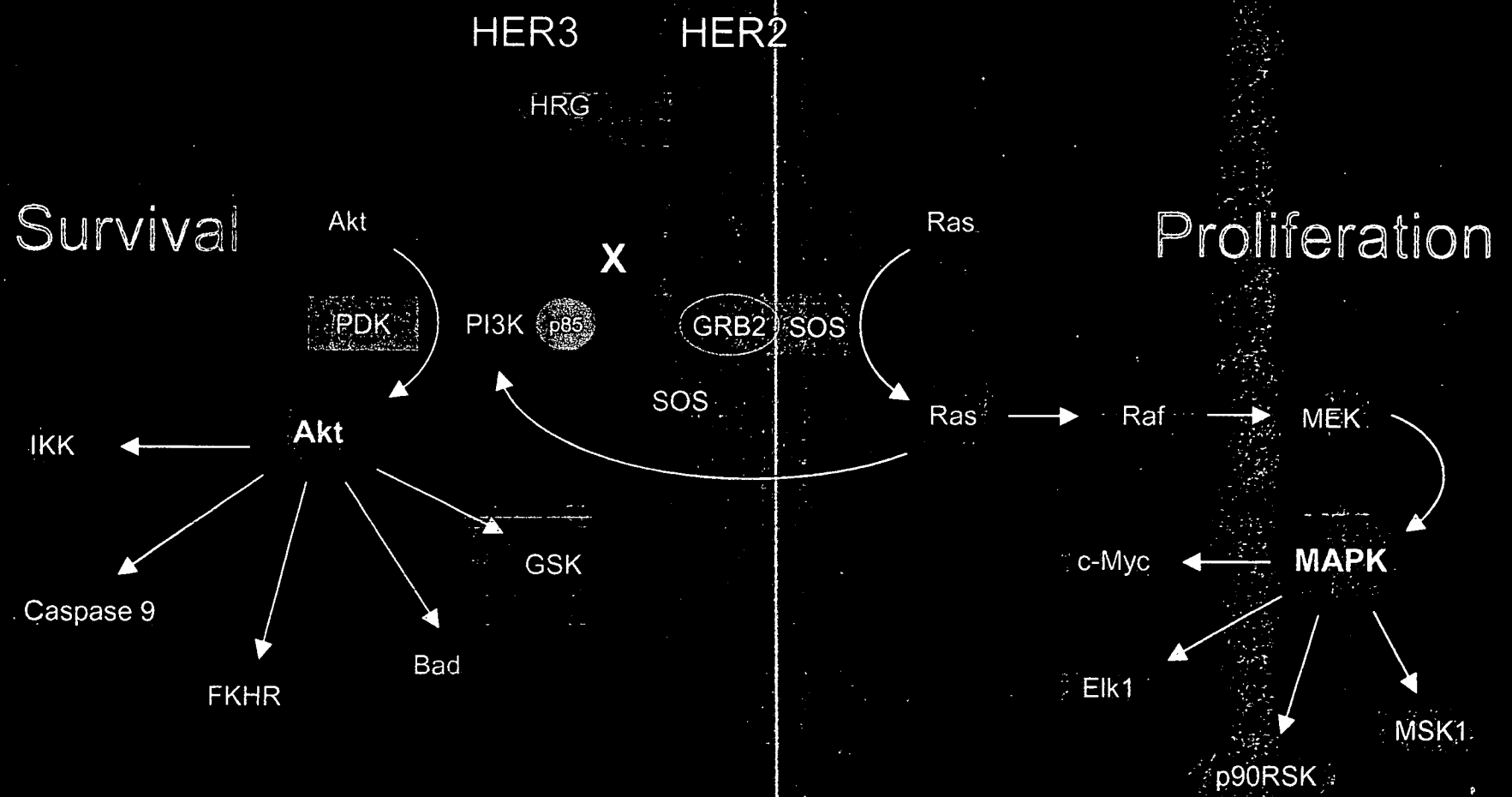


2C4 Inhibits Heregulin-Dependent HER3-HER2 Signaling

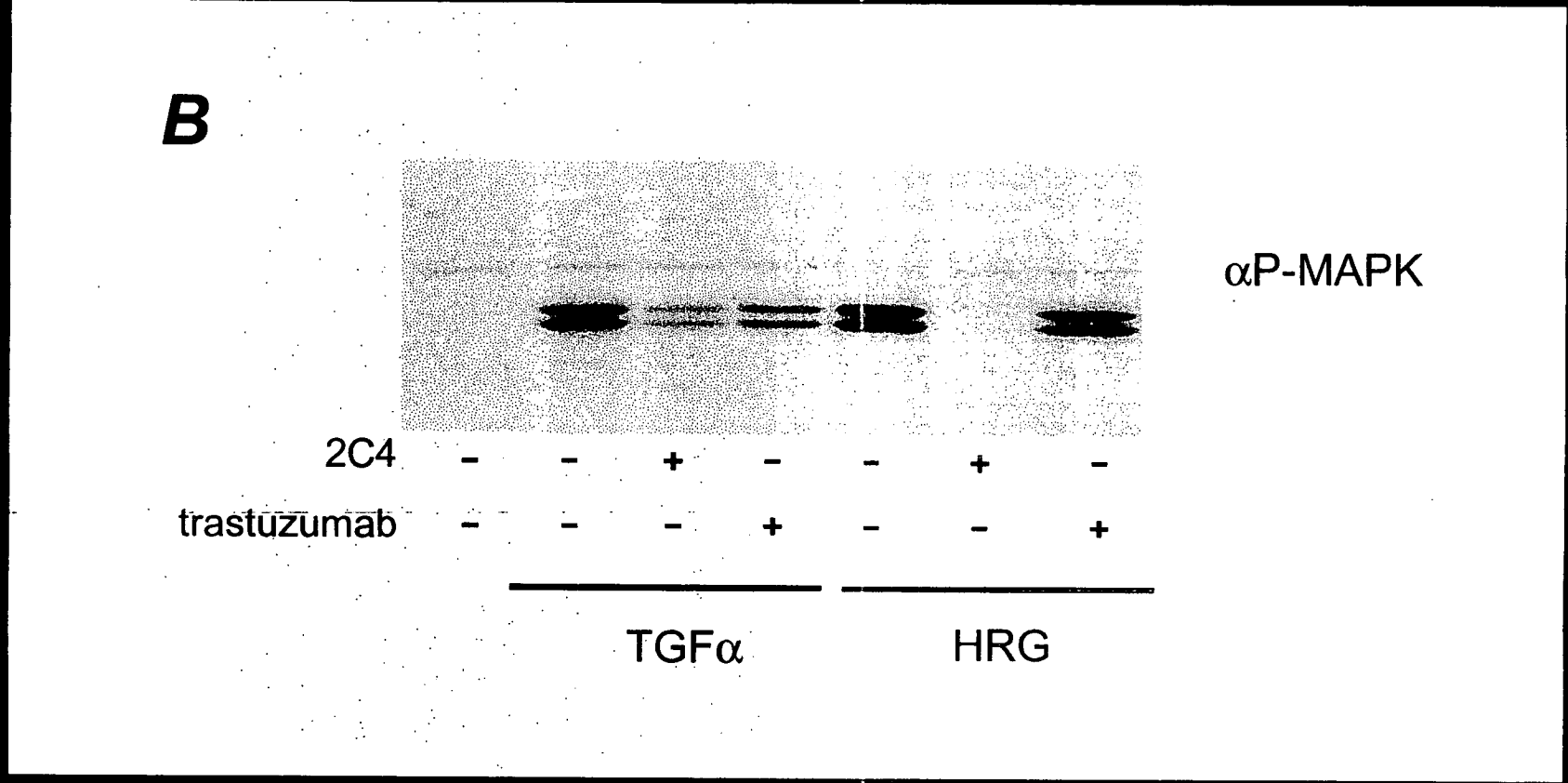


Rob Akita

Coupling of HER2/3 to the MAPK and Akt Pathways

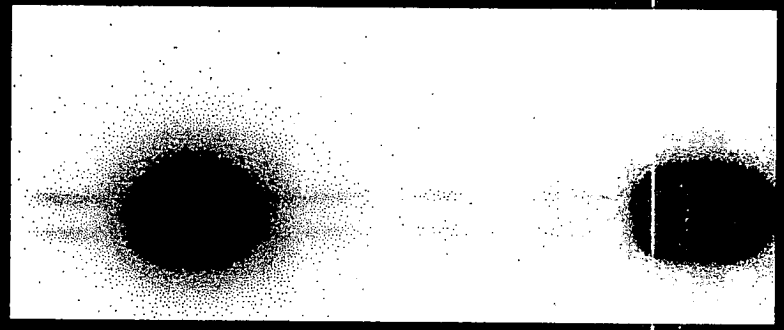


2C4 Inhibits Ligand-Dependent HER2 Signaling (MAPK)



Julie Lofgren

2C4 Inhibits Heregulin-Dependent Akt Activation



GSK3 α/β →

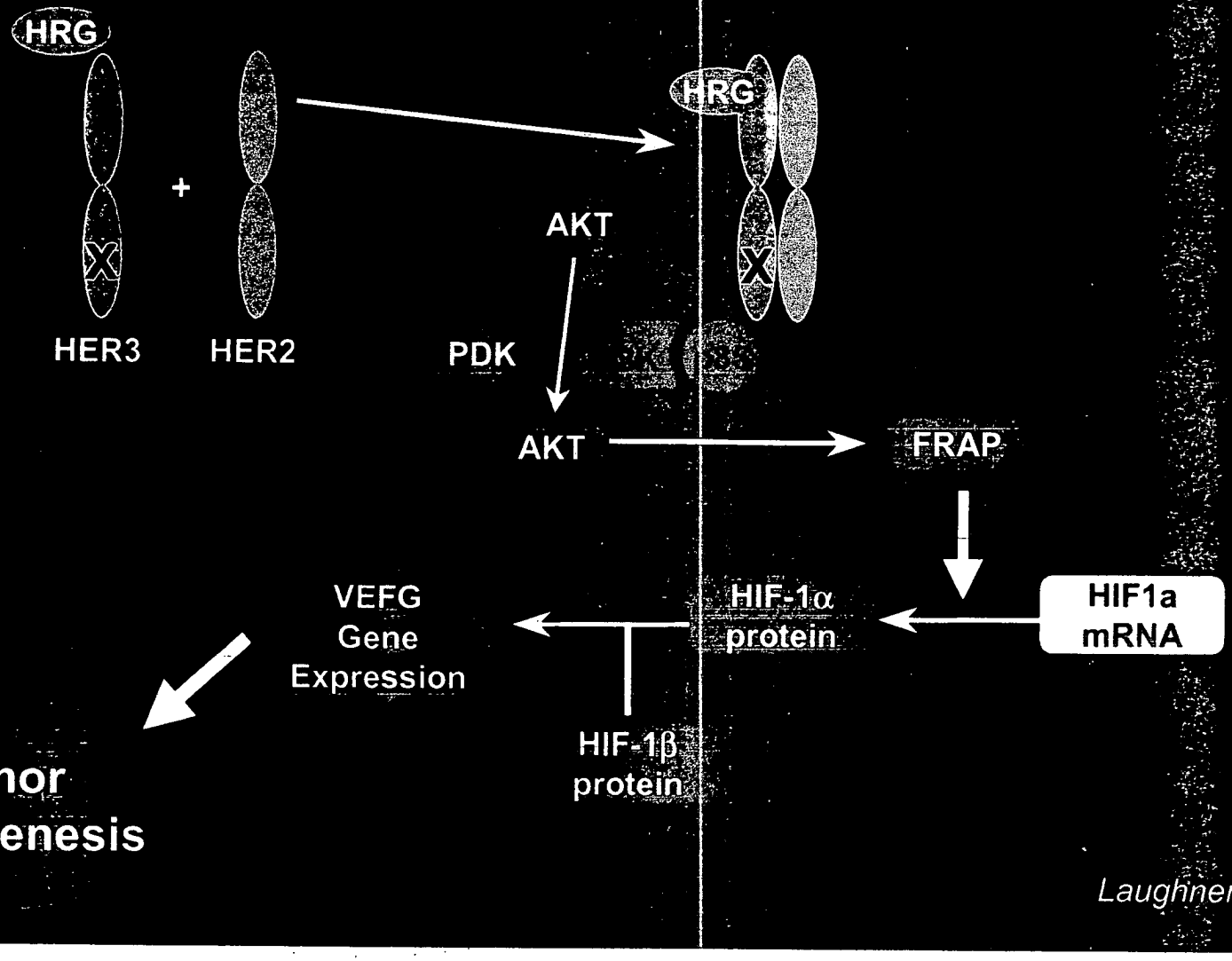
HRG	-	+	-	+	-	+
2C4	-	-	+	+	-	-
Herceptin	-	-	-	-	+	+

ErbB3

1035 pY5
1178 pY5
1180 grb7
1203/05 pY5
1241 pY5
1243 grb7
1257 pY5
1270 pY5
1309 shc

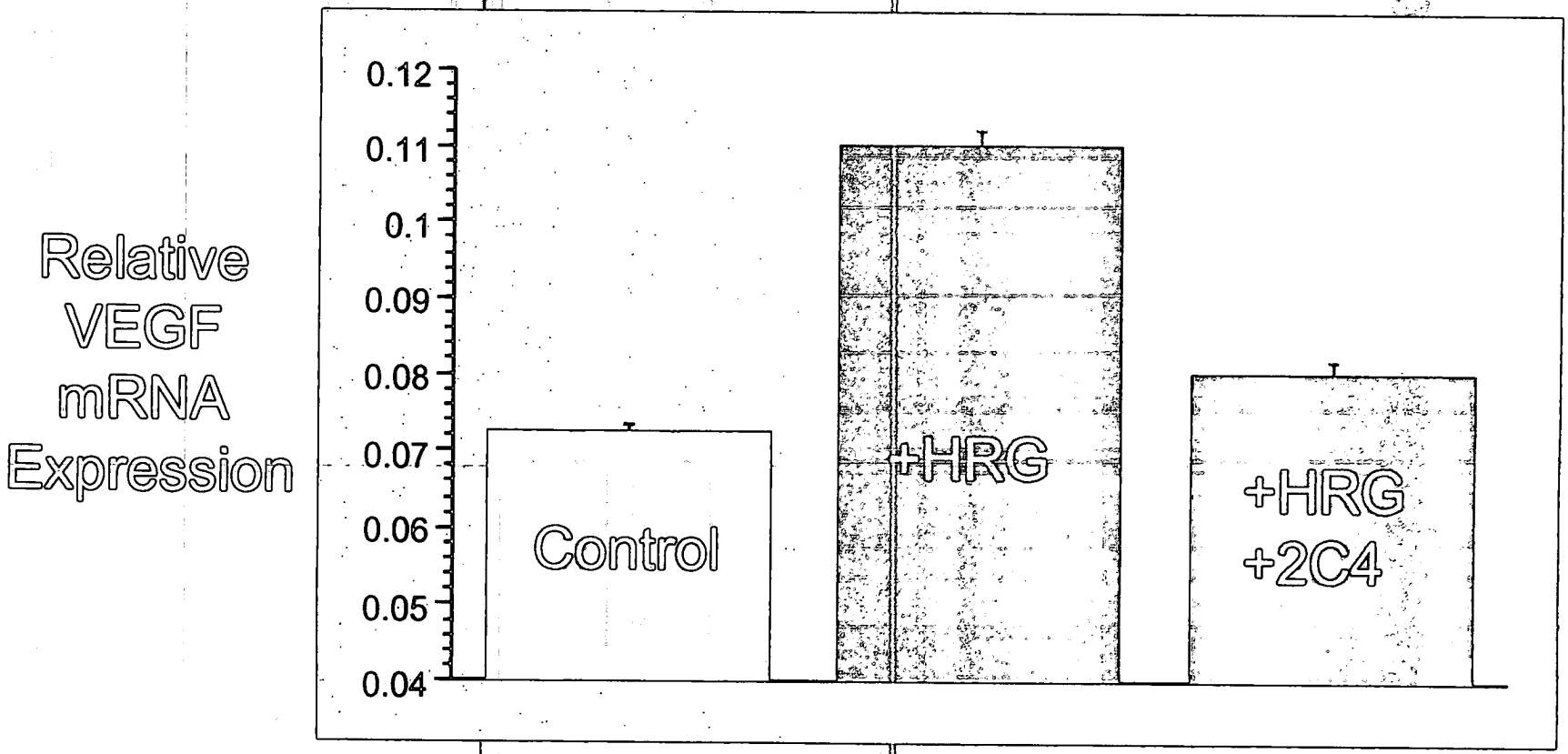
Rob Akita

HER2/HER3 receptor activation increases the rate of hypoxia-inducible factor (HIF-1α) synthesis



Laughner et al. MCB, 2001

2C4 Blocks Heregulin-Induced Expression of VEGF



P. Pisacane

Genentech Acknowledgments

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Lisa Crocker
Gail Phillips
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Len Presta
Matt Franklin
Ken Carey
Bart de Vos

Prostate Cancer and HER2

- Clinical studies:

  HER2 gene amplification or protein overexpression is rare.

  HER ligand expression (e.g., TGF-α) frequently occurs with the onset of the androgen-independent phenotype.

Prostate Cancer and HER2

- Laboratory studies:

  Onset of the androgen-independent phenotype corresponds with HER2 overexpression. (Sawyers).

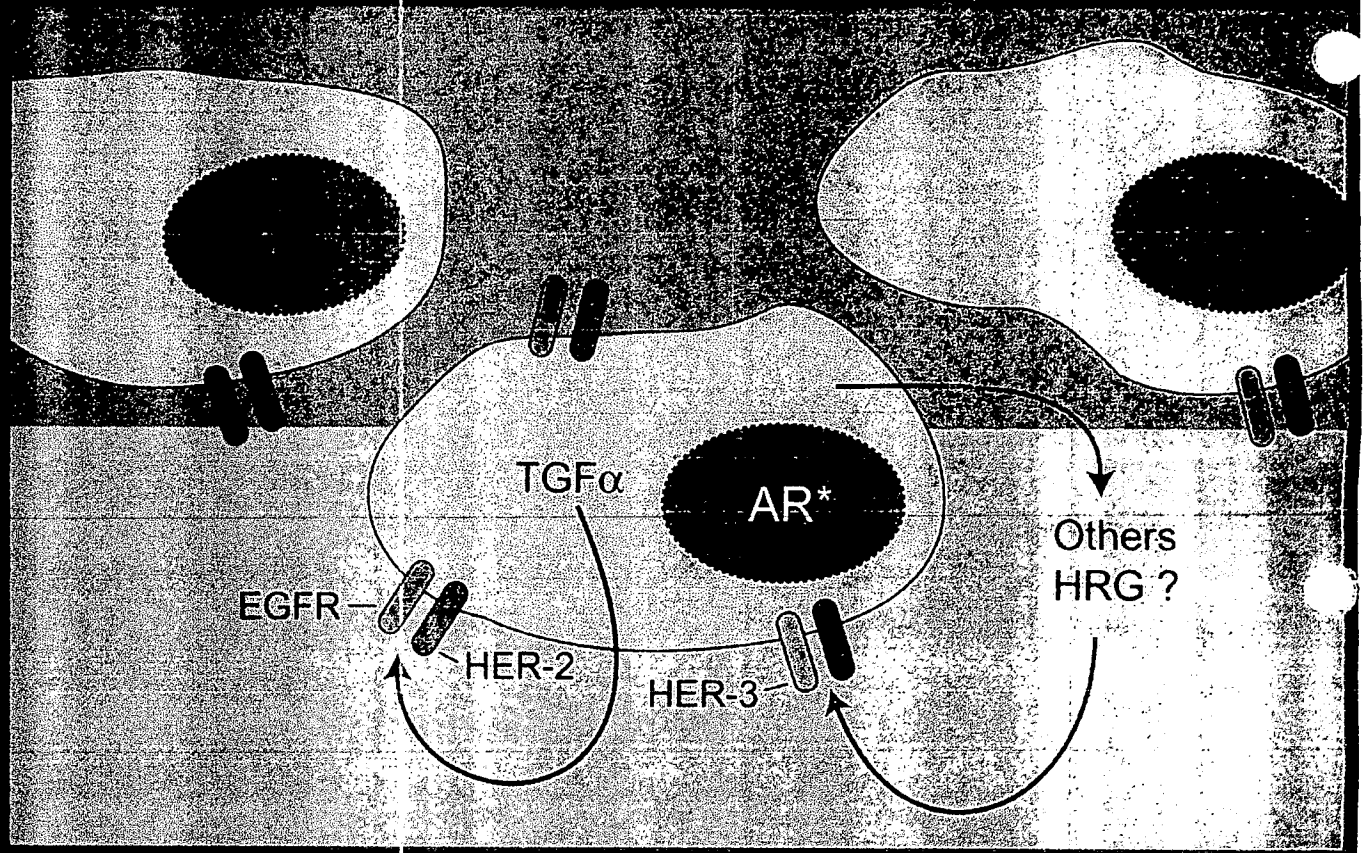
  Evidence for cross-talk between HER2 and androgen receptor signal transduction pathways (Chung).

Androgen–Independent Prostate Cancer

Autocrine activation of HER-kinase axis

Dysregualtion of AR; *unresponsive* to androgen ablation

Increased expression of HER2?

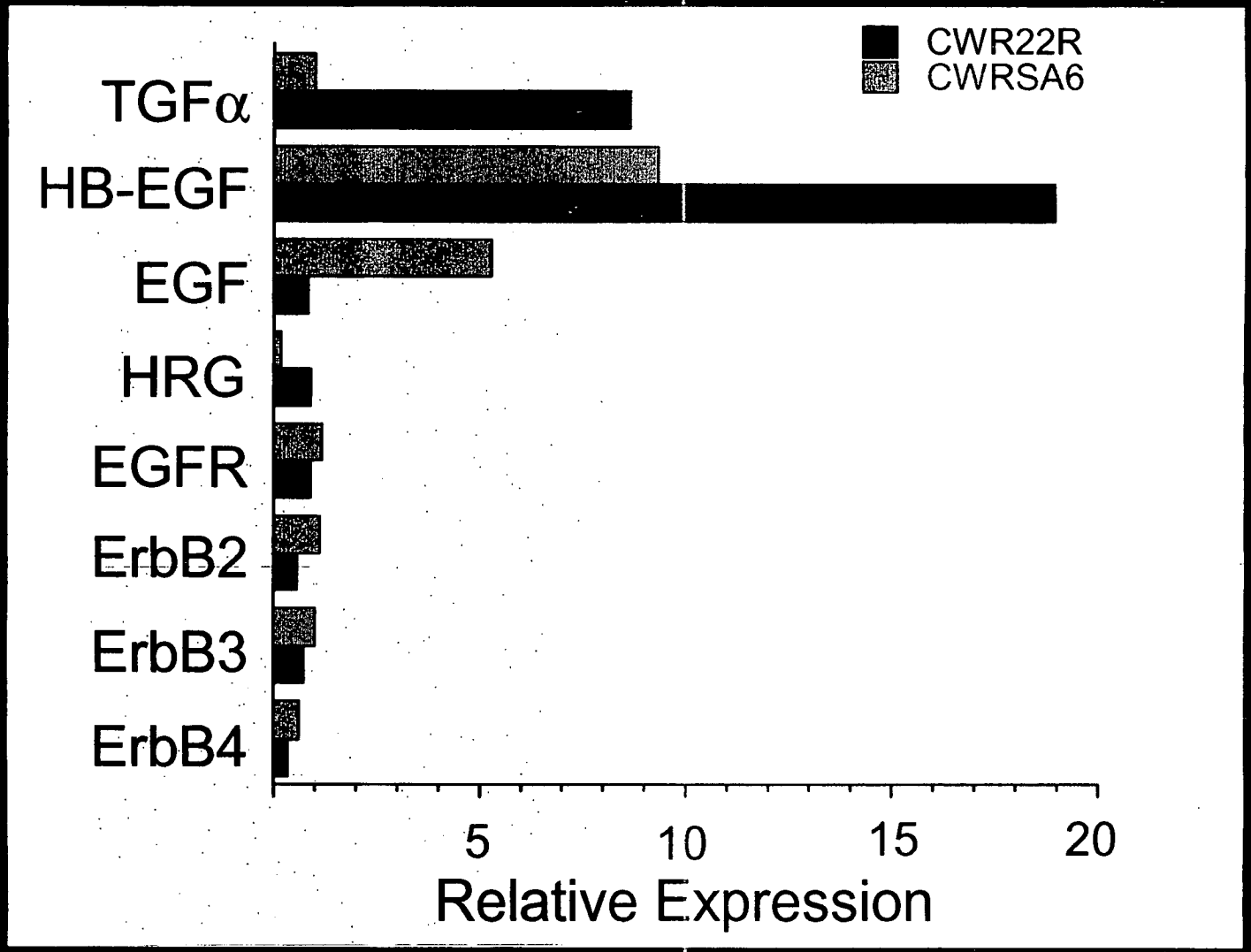


adapted from Kim et.al. (1999)

CWR Prostate Cancer Models

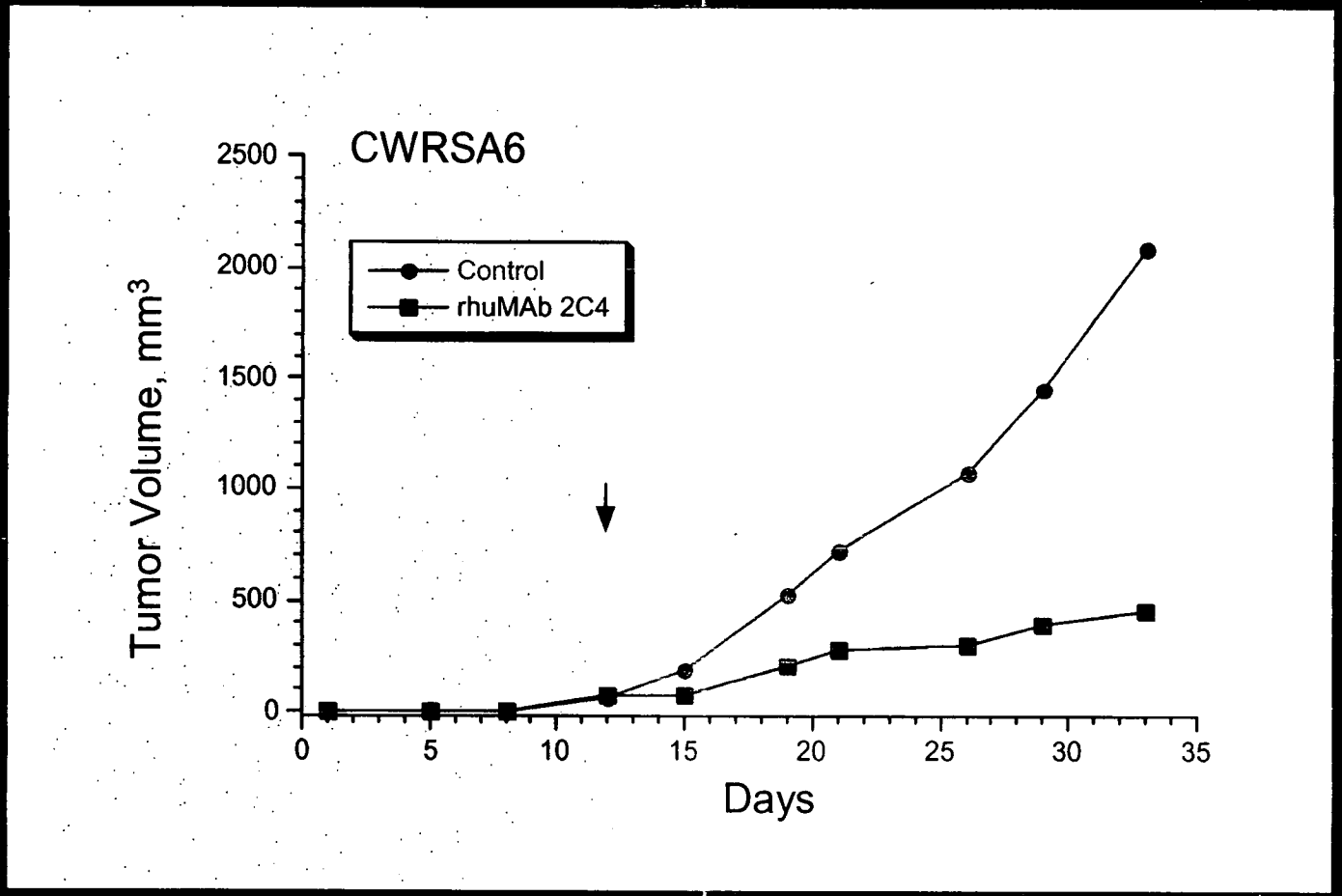
- Derived from a primary prostate cancer patient by Thomas Pretlow, Case Western Reserve.
  - Xenograft maintained by serial transplantation in nude mice.
  - Growth is androgen-dependent (CWR22).
  - Good correlation between tumor growth and serum PSA levels.
- Tumors regress after androgen withdraw.
  - Relapsed tumors are androgen-independent (CWR22R & CWRSA6).

Relative Expression of ErbB Receptors and Ligands in CWR Tumors

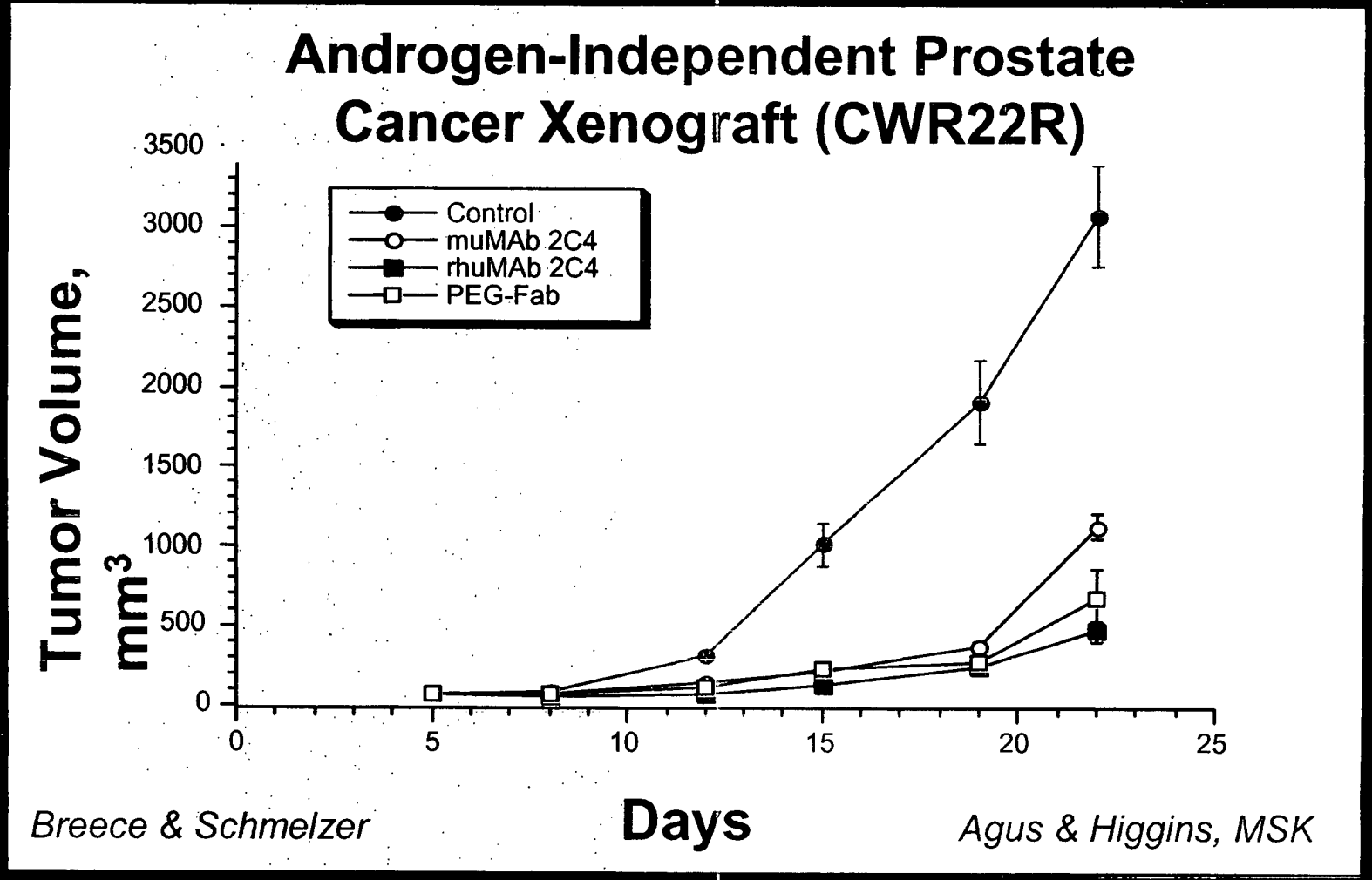


P. Pisacane

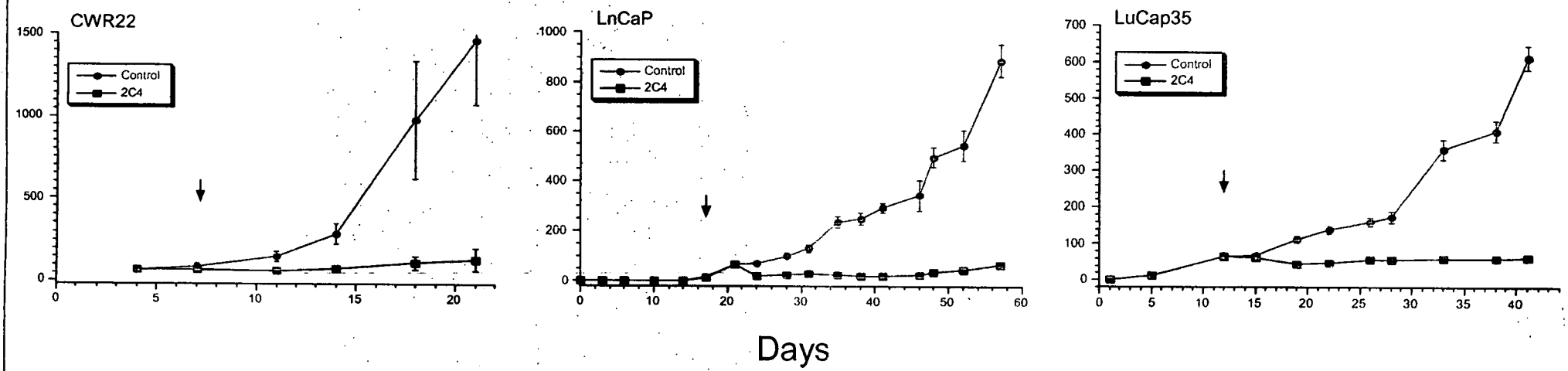
Effect of 2C4 on the Growth of the Androgen-Independent Human Prostate Cancer Xenograft CWRSA6



Proof of Concept Experiment: 2C4 Does Not Require An Intact Fc For Anti-Tumor Activity



Effect of 2C4 on the Growth of the Androgen-Dependent Human Prostate Cancer Models

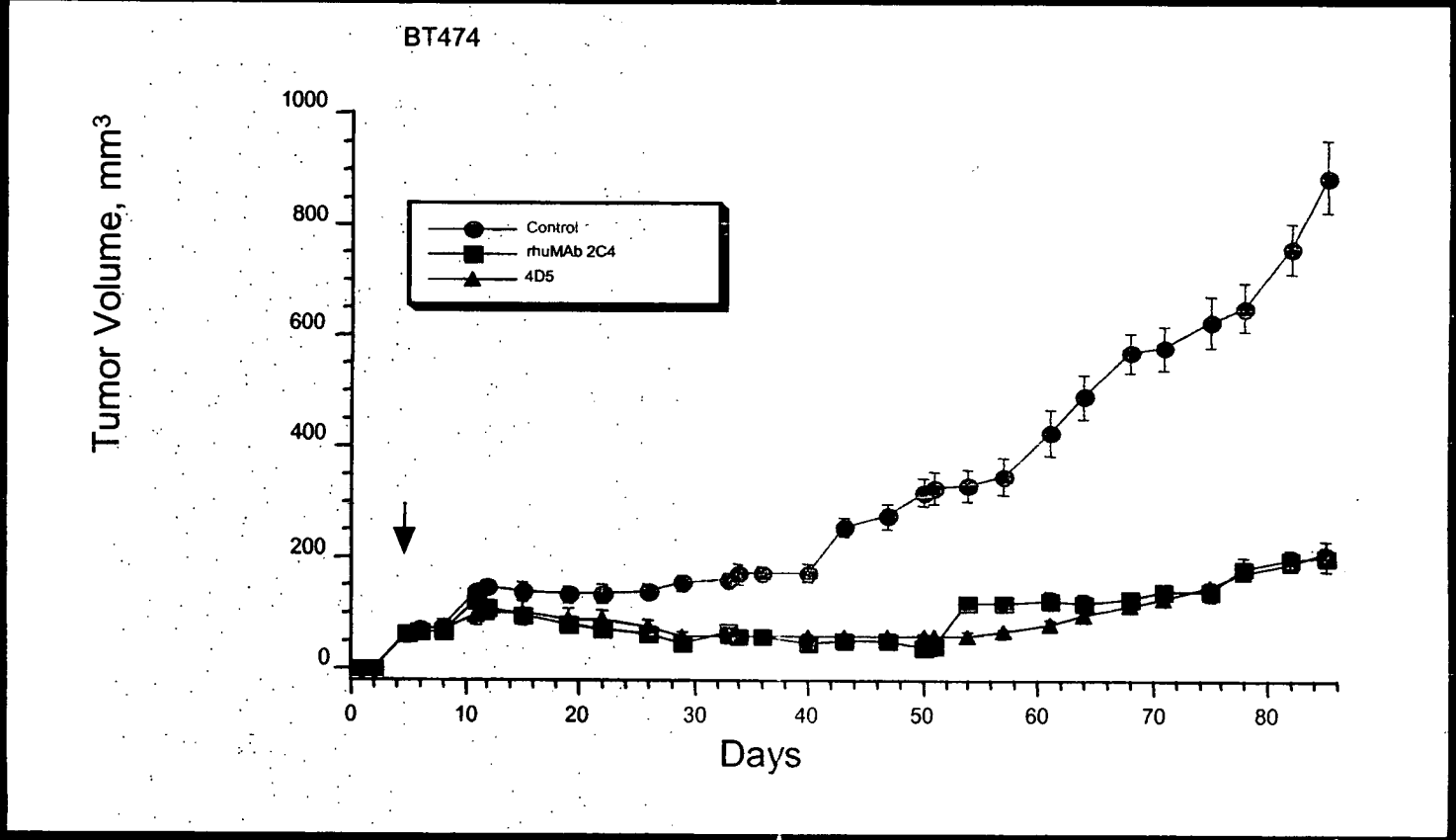


Summary of prostate cancer studies

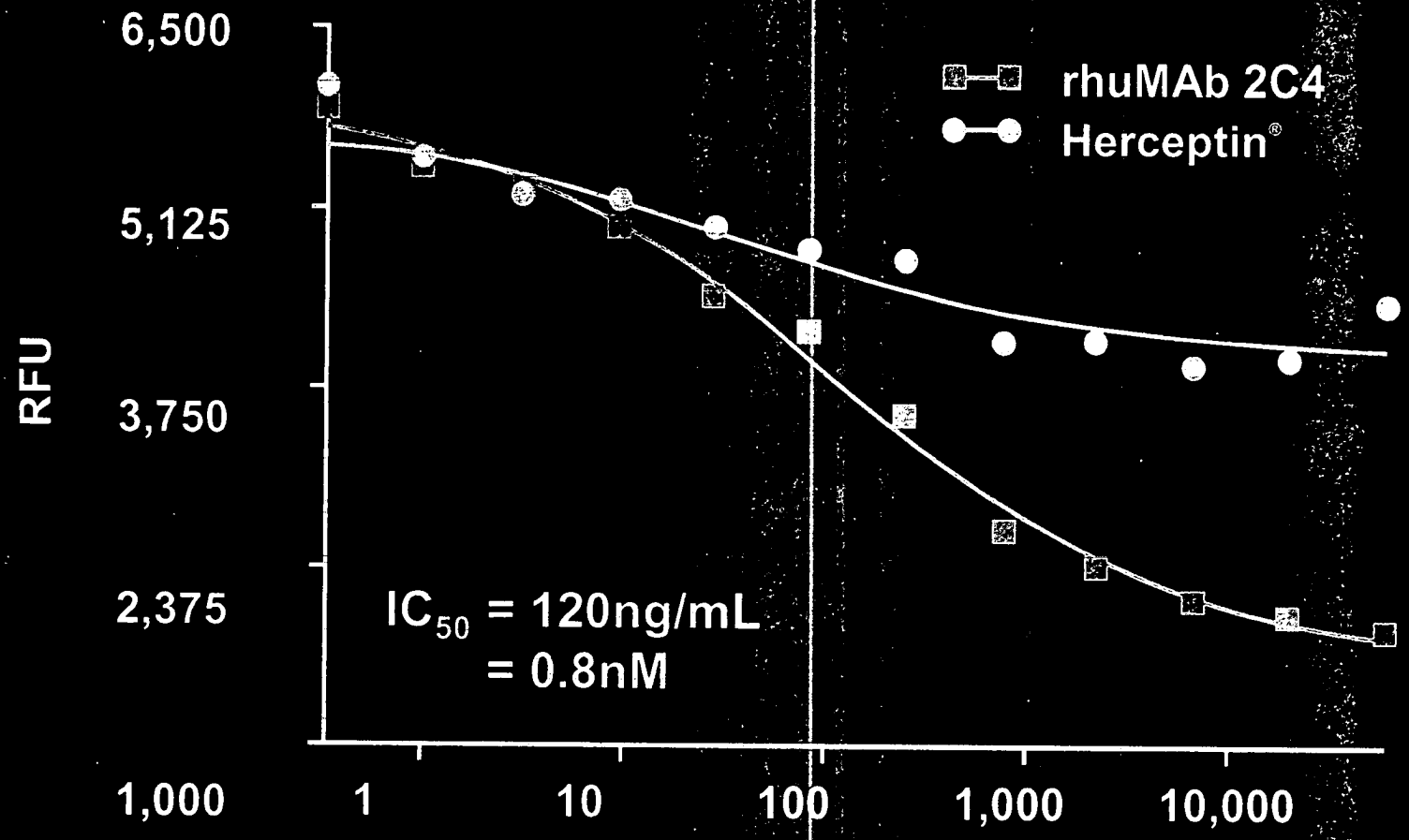
- In contrast to Herceptin®, 2C4 inhibits the growth of androgen-independent prostate tumor xenografts
  - model represents a patient population that is readily available for clinical studies
- Combing 2C4 with low-dose Taxol® results in significant tumor regression and in many cases tumor elimination
- 2C4 also inhibits the growth of androgen-dependent prostate tumor xenografts. These data suggest that 2C4 may be active in patients with early-stage prostate cancer

Breast Cancer Studies

2C4 Has Herceptin-Like Activity Against High HER2 Expressing Tumors



Effect of rhuMAb 2C4 or Herceptin® on the growth of human breast cancer cells (low HER2 expression)

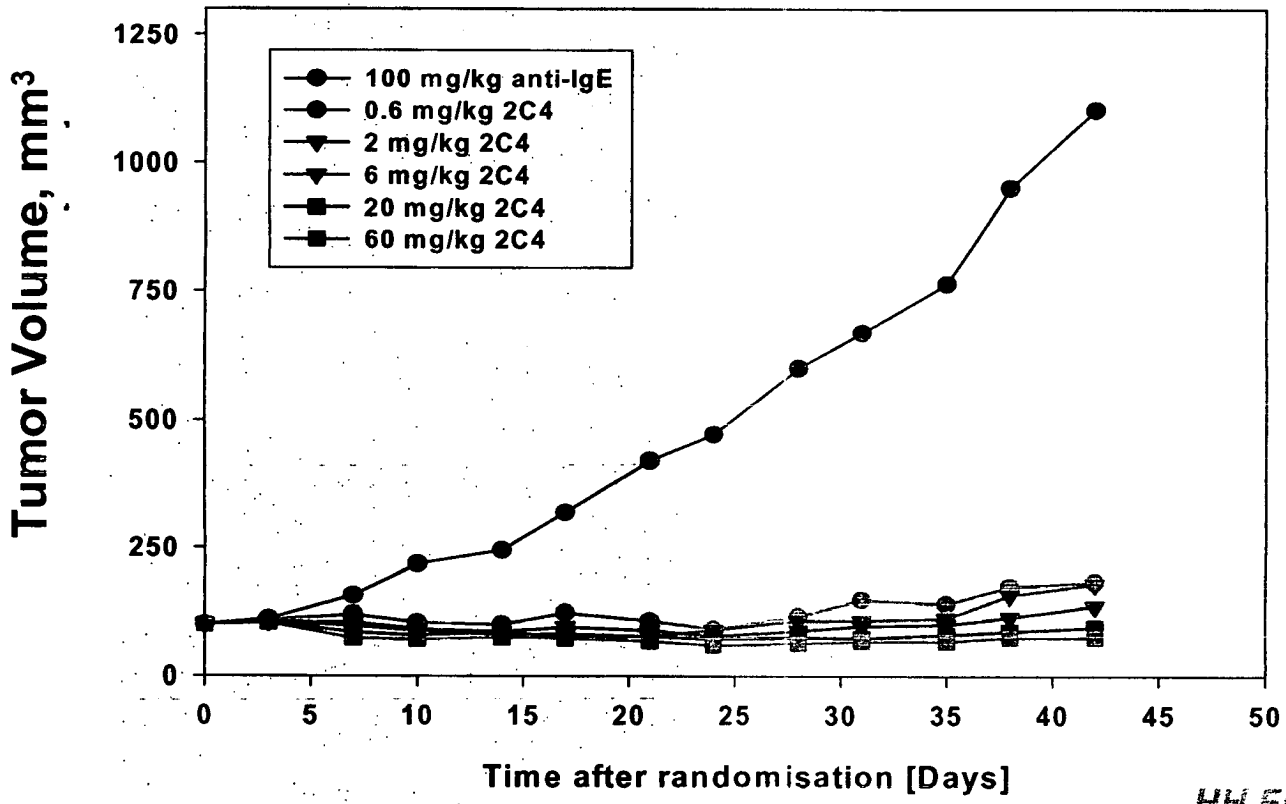


–3 day assay
(Alamar Blue)

K. Totpal

Evaluation of rhuMAb 2C4 in the breast cancer xenograft MAXF 449 (low HER2 expression)

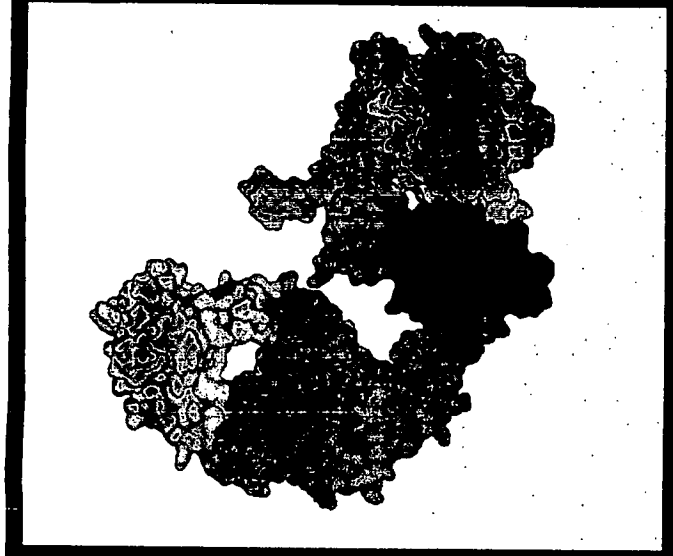
Treatment schedule: i.p.; once/week (Day 1, 8, 15, 22, 29 and 36; 2x loading dose at day 1)



HH Fiebig, Oncotest

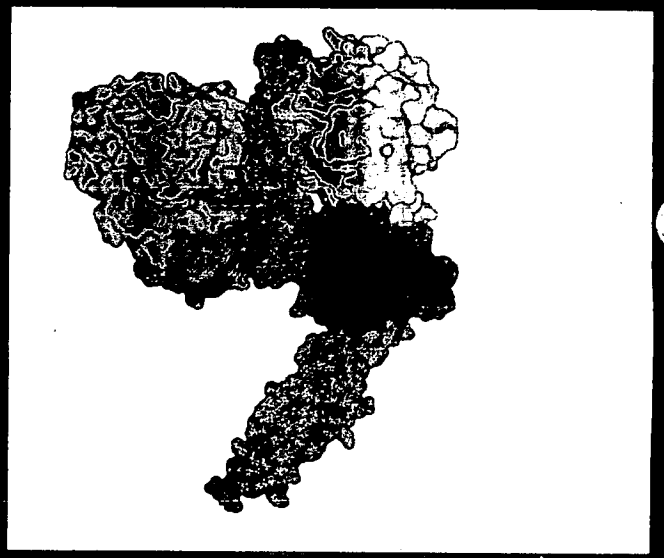
Properties of rhuMAb 2C4

Trastuzumab
Herceptin



- Binds in IV near JM.
- Protects against receptor shedding.
- Moderately affects receptor down-modulation.
- Slight effect on HER2's role as a coreceptor.

Pertuzumab
2C4



- Binds in II at dimerization interface.
- Does not prevent receptor shedding.
- Moderately affects receptor down-modulation.
- Major effect on HER2's role as a coreceptor.

Collaborators

- David Agus: Cedars Sinai
- Howard Scher: Memorial Sloan-Kettering
- Hans-Joachim Mueller: Roche-Penzberg
- HH Fiebig: Oncotest Freiburg